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The International Bureau transmits herewith the following documents and number thereof:

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(54) Title: A PROCESS FOR THE SIMULTANEOUS PRODUCTION OF XYLITOL AND ETHANOL

(57) Abstract

The invention relates to a process for the simultaneous production of xylitol and ethanol from a hydrolyzed lignocellulose-containing material, wherein the starting material is fermented with a yeast strain, the ethanol produced is recovered, a chromatographic separation is carried out on the remaining xylitol solution, and pure xylitol is crystallized.

A process for the simultaneous production of xylitol and ethanol

The present invention relates to a process for the simultaneous production of xylitol and ethanol. A hydrolyzed lignocellulose-containing material is used as a starting material, and in accordance with the process the starting material is fermented with a yeast strain, whereafter the ethanol is recovered and a chromatographic separation is carried out on the fermented solution to obtain pure xylitol.

Xylitol is a naturally occurring sugar alcohol which is formed in the reduction reaction of xylose and which corresponds to "normal" sugar in sweetness and caloric content (4 kcal/g). Xylitol is found in small quantities in many fruits and vegetables and is also produced in the human body as a normal metabolic product. Xylitol is a very good special sweetener in different connections on account of its certain metabolic, dental and technical properties. It may be mentioned by way of example that xylitol metabolism is independent of the insulin metabolism, and therefore also diabetics can use xylitol. Xylitol also has a retarding effect on the bowel, wherefore it may have utility in reducing diets. Furthermore, it has been found that xylitol does not cause caries but has a cariostatic effect.

Despite the many advantages of xylitol, its use has been rather restricted. The reason for this is the relatively high price of xylitol, which in turn is a result of the difficulties of producing xylitol on a larger scale.

Ethanol is a well-known compound which has a wide use.

Xylitol has earlier been produced from xylane-

containing materials by hydrolyzation, in which process a monosaccharide mixture containing e.g. xylose is obtained. Xylose is then converted to xylitol, generally in the presence of a nickel catalyst, such as Raney nickel. A number of processes for the production of xylose and/or xylitol from a xylane-containing material have been described in the literature in this field. As examples may be mentioned U.S. Patent 3 784 408 (Jaffe et al.), U.S. Patent 4 066 711 (Melaja et al.), U.S. Patent 4 075 406 (Melaja et al.), U.S. Patent 4 008 285 (Melaja et al.) and U.S. Patent 3 586 537 (Steiner et al.).

These prior processes are all multi-step processes which are relatively costly and have inadequate efficiency. The greatest problems reside in the effective and total separation of xylose and/or xylitol from polyols and other hydrolysis by-products and the use of the by-products which are produced in large quantities in the process. The purification is very exacting for instance on account of the fact that the catalysts used in the reduction reaction of xylose are very sensitive. The purity of the final product for its part is greatly dependent on that the xylitol can be separated from the other products produced in the reduction reaction.

It is known that several yeast strains produce reductase enzymes which catalyze the reduction of sugars into corresponding sugar alcohols. Certain Candida strains have been reported to produce xylitol from xylose (Ditzelmuller, G. et al.: *FEMS Microbiology Letters* 25 (1985), pp. 195 - 198, Kitpreechavanich, M. et al.: *Biotechnology Letters* Vol. 6 (1984), pp. 651 - 656, Gong, C-S. et al.: *Biotechnology Letters* Vol. 3 (1981), pp. 125 - 130). However, these studies have been carried out on a laboratory

scale only, and the literature in this field has not disclosed processes wherein crystalline pure xylitol is separated from the fermentation product.

The Applicants' copending U.S. application 297 791, filed on January 17, 1989, describes a process for the production of pure crystalline xylitol from plant material using chromatographic separation following hydrolysis and fermentation. However, in this process the majority of the raw material is lost as a worthless waste material. If a greater part of the raw materials could be converted to commercial products, this would essentially improve the economy of the overall process.

It is known that ethanol can be produced from cellulose and hemicellulose by fermenting with a suitable yeast strain. The production of ethanol from D-xylose has been described for instance in U.S. Patent 4 368 268 (C-S. Gong), which publication particularly relates to the manufacturing of mutants which produce ethanol in high yields, and in *Biotechnology and Bioengineering Symp. 12* (1982), pp. 91-102, McCracken, L. & Gong, C-S., wherein fermentation is performed with thermotolerant yeasts.

It has now been found that xylitol and ethanol can be produced simultaneously by using the process of the invention wherein xylose is converted to xylitol, while the majority of the other hexoses present in the raw material are converted to ethanol. Thus the raw material is effectively utilized and two commercially very important products are obtained in a pure form and with a high yield. The process is simple and effective.

The process of the invention is characterized in that the hydrolyzed starting material is fermented with a yeast strain, the ethanol produced is re-

covered, a chromatographic separation is carried out on the remaining xylitol solution, and pure xylitol is crystallized. Xylose-containing substances are used as starting materials, which in accordance with the invention are fermented with a yeast strain that is capable of converting xylose to xylitol and most hexoses to ethanol. By fermentation, a xylitol-rich solution is obtained wherefrom xylitol is recovered in a simple way. Laborious and complex separation steps (such as the conventional ion exchange, demineralization, precipitations etc.) are not needed, but generally the xylitol can be purified in a single step chromatographically, whereafter it is crystallized to obtain pure xylitol. Ethanol is easy to remove from the fermentation solution for instance by evaporation. Thus the need for separating xylitol from the hexitols and other sugars produced in the hydrolysis and reduction steps is avoided. The hydrolysis performed in accordance with the invention also provides a solution to the problem of using pulp discarded as waste mass in other processes, and thus in the process of the invention substantially the entire starting material is utilized.

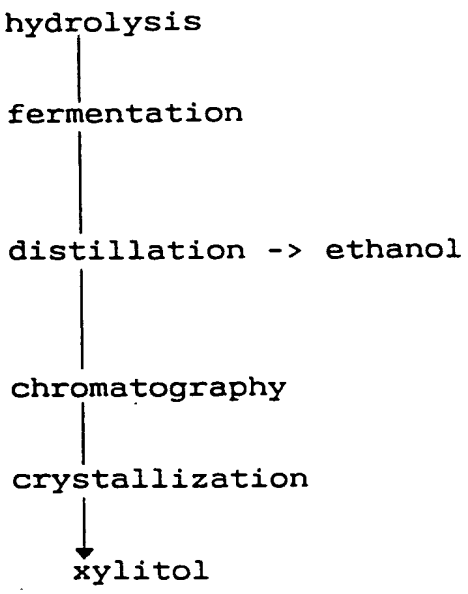
Almost any xylane-containing material can be used as a starting material in the process of the invention. Possible starting materials include softwood, such as birch, beech, poplar, alder etc., and plants or plant constituents, such as straw or hulls of wheat, corn, oat or barley, corn cobs and stems of corn, nutshells, bagasse, and cottonseed bran. When wood is used as a starting material, it is advantageously comminuted or used as chips, sawdust, etc. and treated by hydrolysis or steam explosion and post-hydrolysis, in which connection a carbohydrate material useful in this invention is obtained.

In addition to the above, for instance by-products which are formed in processing and production of woodpulp and which have a high xylane or xylose content can be used. As an example may be mentioned the acid sulphite waste liquor produced in the manufacture of woodpulp by the sulphite process, said waste liquor containing small quantities of undissolved wood solids, and soluble substances such as lignosulphonates, hexoses and pentoses, including xylose, and being a good raw material for use in the production of xylitol. Other by-products and waste products produced in the processing of paper and woodpulp, such as prehydrolysates from the production of viscose mass and waste liquor from the so called neutral sulphite process, which have a high xylane and/or xylose content, can also be used.

The process of the invention employs an aqueous solution containing free xylose. Thus it may be necessary to carry out an acid and/or enzyme hydrolysis on the starting material to break down the xylane into xylose. Processes for hydrolyzing xylane-containing materials to produce xylose-containing solutions have been described e.g. in U.S. Patents 3 784 408 (Jaffe et al.) and 3 586 537 (Steiner et al.).

The starting material may, if desired, be pretreated before the fermentation to remove constituents which may be toxic or otherwise disadvantageous to the yeast. The necessity of the pretreatment step is dependent on the starting material used and the yeast used in the fermentation step. The pretreatment of the starting material may include for instance posthydrolysis, chromatographic separation, ion exchange purification, precipitation, etc.

The process chart is as follows:



The hydrolysis can comprise two steps, prehydrolysis of the cellulose-containing raw material, which may be effected using the so called steam explosion method, and the enzymatic hydrolysis of the polysaccharides and oligosaccharides to produce the corresponding monosaccharides. This step is carried out using enzymes which have a high cellulolytic and xylanolytic activity.

The remaining solids, consisting for the most part of lignin, are then separated from the solution obtained. Alternatively, said solids and the solids produced in the fermentation, such as yeast, can be separated or collected after the next distillation.

When relatively impure solutions are used as a starting material, pretreatment of the solutions may be necessary in some cases. The pretreatment may be e.g. posthydrolysis and/or separation of the constituents which may be toxic and/or disadvantageous to the yeast employed or which have an adverse effect on the fermentation or separation steps. The pretreatment may also be combined with chromatographic sep-

aration, ion exchange purification, precipitation, etc.

Thereafter, the solution is fermented with a suitable yeast strain. The invention employs yeasts that are capable of reducing xylose into xylitol and hexoses into ethanol and/or use hexoses for their growth. Such yeasts are for instance yeasts of the genera Candida, Pichia, Pachysolen and Debaryomyces. Candida and Debaryomyces species, particularly Candida tropicalis and Debaryomyces hansenii, are regarded as advantageous. As a good example may be mentioned the Candida tropicalis strain deposited at the American Type Culture Collection under the accession number ATCC 9968.

The xylose content of the aqueous solution to be fermented is dependent on the starting material and process steps employed, but is advantageously about 50 - 300 g/l.

The fermentation can be carried out in most commercially available fermentors which are furnished with aerating means and stirring and pH regulating means. The temperature is advantageously about 20 - 40°C, most advantageously about 30°C. The yeast cells are added to the xylose-rich solution. Generally, it can be said that the higher the yeast concentration, the faster the fermentation step is. It has been found that the yeast concentration is advantageously about 1 - 20 g of dry yeast/l of substrate (dry weight) when the xylose content is about 50 - 300 g/l.

The fermentation can be enhanced by adding nutrients, and it is continued until the most part of the xylose has been converted to xylitol and substantially all hexoses have been converted to ethanol and/or used for yeast growth. The fermentation gen-

erally takes about 24 - 144 hours, preferably 24 - 72 hours. With the process of the invention, up to 90% of the xylose can be converted to xylitol.

After the fermentation step, the solution is clarified prior to the separation of xylitol and ethanol therefrom. The yeast cells are removed after the fermentation. This may be carried out by centrifugation, filtration or some other similar procedure. When the yeast cells have been removed and the solution is clear, the ethanol produced in the fermentation is recovered by evaporation, distillation or a similar procedure. Alternatively, the removal of the yeast cells can be carried out after the distillation.

To recover xylitol, chromatographic separation is first performed. This is advantageously carried out in a column filled with a sulphonated polystyrene resin cross-linked with divinylbenzene in the alkali/alkaline-earth form. A large-scale chromatographic method suitable for this purpose has been described in U.S. Patent 3 928 193 (Melaja et al.). The chromatographic separation may also be carried out using a simulated mobile bed, as described in U.S. Patent 2 985 589. A DVB cross-linked sulphonated polystyrene resin is used as a filler for the column.

From the fraction having a high xylitol content obtained from the chromatographic step, xylitol can be crystallized with a good yield using conventional crystallization methods, such as cooling or evaporation crystallization. When cooling crystallization is used, xylitol crystals of an average diameter of about 30 µ are added as seed crystals to the concentrated xylitol solution, whereafter the temperature of the solution is slowly decreased. The crystals obtained, the average diameter of which is about

250 - 600 µ, are separated for instance by centrifugation and washed with water to obtain substantially pure crystalline xylitol.

The process can also be carried out in a preferable alternative way so that the starting material is subjected to partial hydrolysis and extraction. The prehydrolysate obtained from the extraction is then fermented to convert xylose to xylitol, which is separated chromatographically and crystallized in the above-stated manner. A final hydrolysis is carried out on the extracted mass, the hydrolysis product is fermented to convert hexoses to ethanol, and ethanol is recovered in the manner described above.

The invention is described in further detail by means of the following examples, which are not intended to restrict the invention.

Example 1

Production of ethanol and xylitol from birch chips

A steam explosion treatment was carried out on birch chips at 215°C with a delay time of 4.5 minutes. The apparatus used is commercially available (Stake Technology, Canada).

30 kg of chips pretreated by steam explosion were suspended in 400 l of water at 50°C in a reactor furnished with stirring means. The pH of the suspension was regulated to 4.8 with a NaOH solution. The following enzymes were added into the reactor:

Cellulase Multifect L 250 (Cultor)	4 FPU/g d.s.
Beta-Glucosidase Novozyme 188 (Novo)	5 IU/g d.s.

Hemicellulase Multifect K (Cultor), containing	xylanase	18 U/g d.s.
	ß-xylosidase	9 nkat/g d.s.
	esterase	2 nkat/g d.s.

The reaction was started, and after three and six hours pretreated birch chips were added to the mixture to increase the solids content to 14% by weight. The hydrolysis was continued for three days at 50°C and at a pH of 4.8. The yield after the hydrolysis was 16% of glucose and 12% of xylose on the dry weight of the pretreated chips.

The solution was separated from the dry solids in a decanting centrifuge (Sharples P 600). The finely powdered matter was removed in a Westfalia Na7-06-076 separator, and the xylose-glucose solution was concentrated by evaporation. The pH of the concentrate was 5.1, and the composition was the following:

glucose	10.3%
xylose	7.6%
other monosaccharides	3.1%
oligosaccharides	5.5%

The total solids content was about 32%.

The solution additionally contained salts of organic acids and small amounts of lignin decomposition products, furfural, phenols and other organic substances.

The hydrolyzed product was fermented with the yeast Candida tropicalis ATCC 9968. A New Brunswick Scientific Co If 250 fermentor was used, whereto gas analysis and mass spectrometric apparatus was connected.

The fermentation solution contained:

60 l	prehydrolysate (dry solids content about 32%)
1.5 kg	Gistex yeast extract (steam sterilized at 121°C, 15 min.)
29 l	water

The inoculation cultures were grown in two stages, first in a 2 l Erlenmeyer flask in an Orbital Shaker at 30°C for 2 days, and then in a Microgen SF 116 laboratory fermentor having an operating volume of 11 l. The fermentor was aerated at a rate of 5.5 Nl/min. (0.5 VVM) and stirred at a rate of 500 rpm. The culturing lasted for one day.

The actual fermentation was performed on a pilot scale, the operating volume being 100 l. The fermentor was aerated at a rate of 20 Nl/min. (0.2 VVM) and stirred at a rate of 100 rpm. The temperature was maintained at 30°C and the pH at 6. Plurior® was used as an antifoaming agent.

The fermentation results have been set forth in Table 1.

Table 1

time (h)	yeast (g/kg)	xylitol (g/l)	glucose (g/l)	ethanol (g/l)
0	2.0	0.0	53.5	1.9
16	6.1	2.9	2.4	26.4
23.5		4.7		26.7
41.0	7.4	9.0	1.9	25.6
65.0	8.0	15.8		24.9
91.5	6.1	21.2		23.4
136		20.6		22.3

After the fermentation, substantially all sugars had converted into xylitol or ethanol.

Ethanol was recovered from the solution by distilling the fermented solution in a conventional manner. The distillation apparatus was constructed of standard components (Corning Process Systems) which were of borosilicate glass, and the apparatus comprised equipment for 15 separation steps as follows: boiler, 13 bubble plates and a feed plate between the fourth and fifth bubble plates seen from the top. The diameter of the column was 10 cm.

The distillation was carried out at a pressure of 110 mbar at a feed rate of 10 l/h and with a reflux ratio of 3:1. 110 l of fermenting solution gave 7.0 kg of distillate which contained 27.1% by weight of ethanol. The ethanol content of the bottom product was 0.02% by weight.

The separation and, if desired, crystallization of xylitol were carried out as described in Examples 2 and 3.

Example 2

Production of ethanol and xylitol from sulphite waste liquor

The starting material used was a sugar fraction chromatographically separated from a sulphite waste liquor (Finnish Patent Application 862273, U.S. Patent 4 631 129), containing a considerable amount of hexoses, mainly glucose. The composition of the solution prior and subsequent to fermentation is shown in Table 2.

Table 2

ingredient	before fermentation	after fermentation
dry solids, % by weight	19.0	-
oligosacch., % of dry solids	14.8	10.3
glucose	90.0	1.4
xylose	42.0	3.5
arabinose	5.0	2.3
xylitol	-	25.4
ethanol	-	42.0
arabinitol	-	2.8

The fermenting was carried out with a Debaryomyces hansenii strain, and 3 g/l of yeast extract, 3 g/l of malt extract and 5 g/l of peptone were added. The pH of the solution to be fermented was initially about 6.0, the temperature was about 30°C and the fermentation was carried out in an Orbital Shaker (200 rpm).

The ethanol produced in the fermentation was recovered by distillation (50°C, 200 mbar), and a chromatographic separation was carried out on the remaining solution in a column filled with a divinylbenzene-cross-linked polystyrene-based cation exchanger, in which connection the following conditions were used:

height of column	4.0	m
diameter of column	22.5	cm
temperature	65	°C
flow rate (H_2O)	30	l/h
feed concentration	30	% by weight

feed volume 6 kg of solid matter
resin: Finex C 09
particle size 0.37 mm
ionic form Na^+

The results have been graphically presented in the figure. Xylitol was separated from xylose and the other impurities, and recovered from the xylitol-rich fraction, wherefrom pure xylitol was crystallized in the manner described in Example 3.

Example 3

Crystallization of xylitol

Xylitol was crystallized from a chromatographically enriched xylitol solution containing 82.5% of xylitol on dry solids by evaporating the solution to 92% by weight of dry solids at 65°C. Into a solution of a natural weight of 2 200 g, xylitol crystals of about 0.04 mm were inoculated in an amount of 0.03% by weight, and the solution was cooled in 55 hours to 45°C in accordance with the following empirical equation:

$T = T1 - (t/t1)^{**}2^* (T1 - T2)$, wherein

T = temperature of solution, °C
T1 = seeding temperature (65°C)
T2 = final temperature (45°C)
t = time from seeding, h
t1 = crystallization time (55 h)

The crystallization was carried out in a 2 l pilot crystallizer furnished with a vertical stirrer. 65% of the xylitol present in the solution crystallized as raw crystals which were separated from the mother solution in a basket centrifuge (Hettich, Roto

Silenta II).

During the centrifugation, the crystals were washed with water (4% of water on the weight of the crystals). The centrifugation time was 5 minutes, and a centrifugal force of 2 000 g was used. 1 510 g of natural weight of a crystal suspension was centrifuged, which gave 705 g of crystalline dry solids having a xylitol content of 99.4% of dry solids. The average size of the crystals was 0.37 mm and the standard deviation 24%.

The raw crystals can be recrystallized into product crystals by the method disclosed in Finnish Patent 69 296.

Example 4

Production of ethanol and xylitol from barley hulls

Barley hull mass having the following carbohydrate composition was used as a starting material:

xylan	21.6% of dry solids
glucan	33.4
arabinan	5.7
galactan	1.4
mannan	0.6
rhamnan	0.2

The barley hull mass was hydrolyzed at a pressure of 350 psi at 235°C, and the delay time was 2.0 minutes. The hydrolyzed material contained 46.6% of dry solids, and the content of dissolved solids was 34.2% on dry solids. The filtrate contained 12.7% of monosaccharides, 16.9% of acetic acid and 0.5% of furfural calculated on dry solids. Posthydrolysis was carried out on the filtrate by adjusting the pH to 1 with sulphuric acid and by hydrolyzing the solution

for 4 hours at a pressure of one atmosphere at 100°C. The composition of the posthydrolysate was the following:

oligosaccharides	1.3% of dry solids		
monosaccharides	45.2%:		
	- xylose	67.3%	of the
	- arabinose	11.4%	mono-
	- glucose	16.0%	sacchar-
	- galactose	3.3%	ides
	- mannose	1.5%	
	- rhamnose	0.5%	
others (e.g. furfural)	3.3% of dry solids		

The fermentation of the posthydrolysate, the recovery of ethanol and the crystallization of xylitol were carried out as described in the preceding examples.

Example 5

Production of ethanol and xylitol from oat hulls

Oat hull mass having the following carbohydrate composition was used as a starting material:

xylan	26.5% of dry solids
glucan	30.7%
arabinan	3.0%
galactan	1.3%
mannan	0.2%

The oat hull mass was hydrolyzed at a pressure of 350 psi at 235°C, and the delay time was 2.0 minutes. The hydrolyzed material contained 39.1% of dry solids, and the content of dissolved solids was 36.4% of dry solids. The filtrate contained 12.0% of mono-

saccharides, 12.9% of acetic acid and 0.5% of furfural calculated on dry solids. Posthydrolysis was performed on the filtrate by adjusting the pH to 1 with sulphuric acid and by hydrolyzing the solution for 4 hours at a pressure of one atmosphere at 100°C. The composition of the posthydrolysate was the following:

oligosaccharides	1.3%	of dry solids
monosaccharides	63.1%:	
- xylose	69.0%	of the
- arabinose	6.9%	mono-
- glucose	19.1%	sacchar-
- galactose	3.1%	ides
- mannose	0.8%	
- rhamnose	1.1%	
others (e.g. furfural)	2.8%	of dry solids

The fermentation of the posthydrolysate, the recovery of ethanol and the crystallization of xylitol were carried out as described in the preceding examples.

Example 6

Steam explosion and extraction of birch chips

A steam explosion treatment was carried out on birch chips with a factory-scale equipment at a temperature of 215°C with a delay time of 4.5 minutes. The manufacturer of the equipment used is Technip, type of apparatus Stake II System.

The steam explosion product was suspended in hot process water in a mixing container to produce a fibrous suspension of about 3.5%. Therefrom the slurry was directed via an overflow to form a smooth layer on a 5-phase band filter operating on the coun-

tercurrent principle (type A 40-B25; manufacturer Filters Philippe; width of wire 2.7 m; wire supplied by manufacturer of apparatus). The solid mass was further extracted with hot water on the wire.

The aqueous solution obtained had:

dry solids content	8.7% by weight
xylose monomers	1.1% of natural weight
xylose oligomers	3.7% "
glucose	0.04% "

Example 7

Enzymatic degradation of steam-exploded water-washed birch chip mass

The composition of the steam-exploded (215°C/4.5 min.) birch chip mass (prepared in accordance with Example 6) used as raw material for the hydrolysis was the following:

dry solids	32%
cellulose	60% of dry solids
xylan	3.6% "
lignin	25% "
(extractable in acetone)	
Klason lignin	12.3% "

90 kg of the above-described mass was weighed into a reaction vessel provided with a stirrer and a heating jacket and containing 370 l of water. The mixture was heated to 50°C, the pH was adjusted to 4.8 - 5.0, whereafter the enzyme solutions were added (1.24 l of Multifect L 250, 0.11 l of Novozyme 188 and 0.09 l of Multifect K). As activity units, the added quantities correspond to 6 FPU/g of cellulase, 5 IU/g of ß-glucosidase and 0.02 ml of growth solution/g of mass dry solids of hemicellulase (18 U/g of dry solids of xylanase, 9 nkat/g of dry solids of ß-xylosidase, 2 nkat/g of dry solids of esterase). The

reaction was allowed to continue under the conditions described above for 18 hours. Thereafter mass and enzymes were added in the same quantities as in the starting phase. A corresponding mass and enzyme addition was repeated after 21 hours from the start. Thereafter the hydrolysis reaction was allowed to continue so that the total time was 40 hours. The enzyme action was then stopped by heating the mass mixture to 80°C for 10 - 20 minutes. In that connection, the remaining solid matter was solidified and thereby made easier to separate. The solid matter and the solution were separated from one another by centrifugation (Pennvalt Sharples P 600 model). The solution was further clarified by separating the remaining fine precipitate in a separator (Westfalia model NA7-06-076). The solution was concentrated to 33% for fermentation by evaporating with a Luwa evaporator in vacuo at a temperature of 40 - 50°C.

Hydrolysis yields of steam exploded, water washed birch chip mass in enzyme treatment:

	% in solution	yield % of dry solids	conversion %
glucose	3.3	24.5	40.8
xylose	0.4	2.6	72.0
oligosaccharides	0.7		

Composition of the clarified and evaporated enzyme hydrolysate solution:

glucose	22.7%	of natural weight
xylose	2.7%	"
oligosaccharides	4.7%	"

Example 8

Fermentation of enzymatic hydrolysate of steam exploded, water washed birch chip mass into ethanol

The hydrolyzed cellulose was fermented with a yeast Candida tropicalis ATCC 9968. A New Brunswick Scientific IF-250 fermentor was used.

The fermentation solution contained:

45	l	hydrolysate
1.5	kg	Gistex yeast extract
40	l	water

The inoculation cultures were grown in two steps, first in a 2 l Erlenmeyer flask in an Orbital Shaker at 30°C for 2 days, then in a New Brunswick Scientific SF-116 laboratory fermentor having an operating volume of 11 l. The fermentor was aerated 5.5 Nl/min. (0.5 vvm) and stirred at a rate of 500 rpm. The culturing lasted for one day.

The actual fermentation was carried out on a pilot scale, the operating volume being 100 l. The fermentor was aerated 25 Nl/min (0.25 vvm) and stirred at a rate of 100 rpm. The temperature was adjusted to 30°C, and the foam was controlled with Plurior antifoaming agent.

The results of the fermentation are set forth in Table 4.

Table 4

time (h)	cell mass (g/l)	glucose (g/l)	ethanol (g/l)
0	1.8	105.0	1.9
19.5	11.3	0	51.2
52	-	0	48.1
66	-	0	45.0

In the course of 29.5 hours, the yeast consumed

all of the glucose in the substrate, producing ethanol therefrom with a yield of 48%.

After fermentation, the yeast cells were separated from the solution by centrifugation (Westfalia NA7-06-076). The clarified solution was distilled to recover the ethanol.

Example 9

Recovery of ethanol from the fermentation product of enzymatic hydrolysate of steam exploded, water washed birch chip mass

100 litres of fermented cellulose hydrolysate were distilled. The fermentation had been carried out in the manner described in Example 8 and clarified by centrifugation in a Westfalia NA7-06-076 separator. The ethanol content of the solution was 3.4%.

The distillation apparatus was constructed of standard components by Corning Process Systems which were of borosilicate glass. The diameter of the column was 10 cm. The apparatus comprised 15 separation steps: boiler, 13 bubble plates and a feed plate between the fourth and fifth bubble plates seen from the top. The distillation was carried out at a pressure of 100 mbar, at a feed rate of 10 l/h and with a reflux ratio of 3:1. 8.5 kg of distillate were recovered, having an ethanol content of 36.0%. The ethanol content of the bottom product was 0.1%.

Claims:

1. A process for the simultaneous production of xylitol and ethanol from a hydrolyzed lignocellulose-containing material, characterized in that the starting material is fermented with a yeast strain which is capable of converting free xylose to xylitol and the free hexoses present to ethanol and yeast, the ethanol produced is recovered and xylitol is chromatographically separated from the remaining xylitol solution.

2. A process according to Claim 1, characterized in that the starting material is extracted, the extracted solution is fermented to convert xylose into xylitol and a chromatographic separation and crystallization are carried out on the xylitol solution and a final hydrolysis is carried out on the extracted mass, said mass is fermented and the ethanol produced is recovered.

3. A process according to Claim 1, characterized in that a xylane-containing lignocellulose, such as birch or grain hulls, is used as a starting material.

4. A process according to Claim 1, characterized in that sulphite waste liquor is used as a starting material.

5. A process according to Claim 1, characterized in that pure xylitol is crystallized from the xylitol-rich fraction obtained in the chromatography step.

6. A process according to Claim 1, characterized in that the yeast cells are removed prior or subsequent to the distillation.

7. A process according to Claim 1, characterized in that the yeast strain is of the

genus Candida or Debaryomyces.

8. A process according to Claim 1 or 7, characterized in that the yeast is a Candida tropicalis species and is preferably Candida tropicalis ATCC 9968.

9. A process according to Claim 1, characterized in that the yeast is a Debaryomyces hansenii species.

10. A process according to Claim 1, characterized in that the ethanol is recovered by distillation.

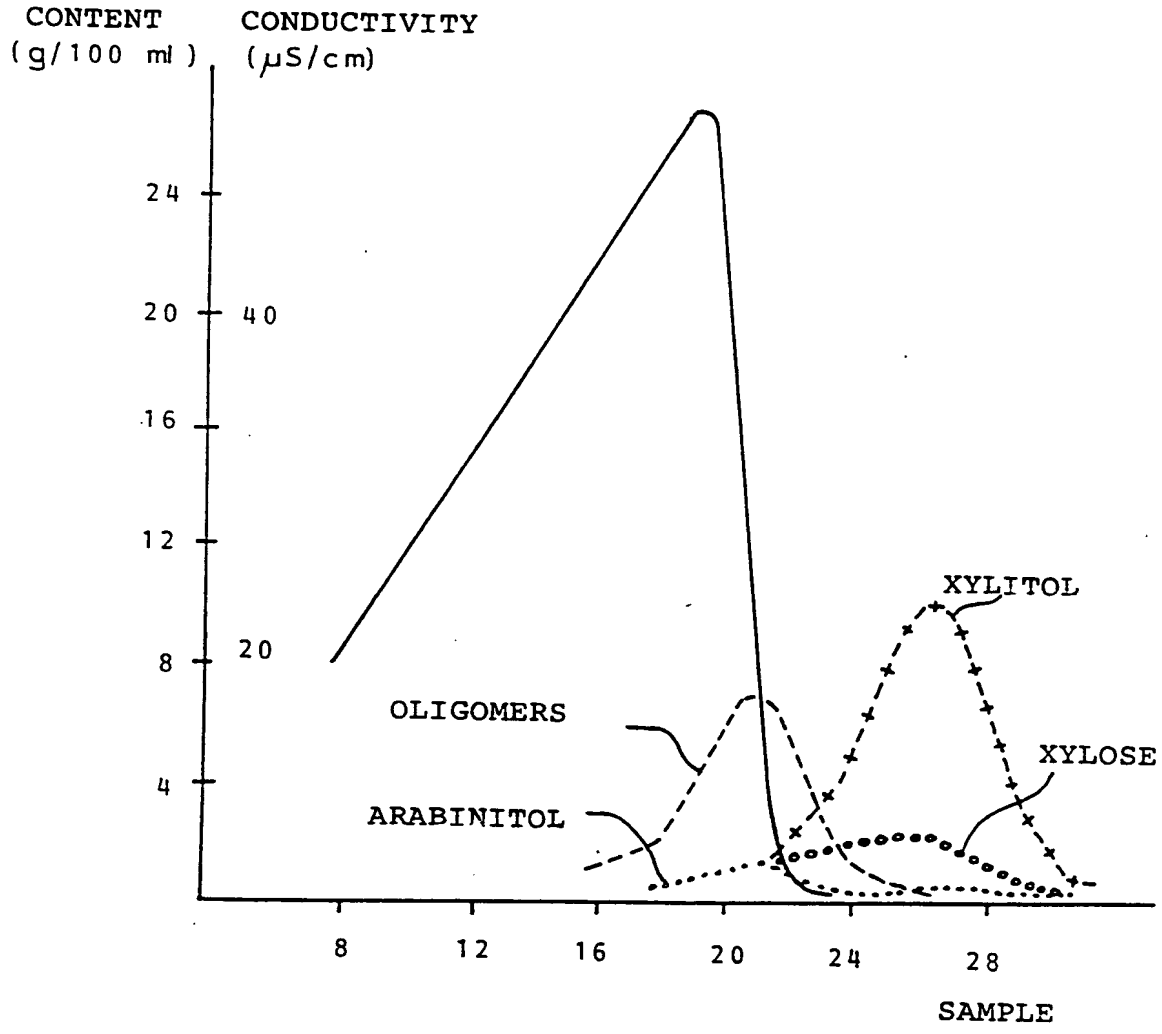
11. A process according to Claim 1, characterized in that the hydrolysis is carried out by steam explosion and enzymatic final hydrolysis.

12. A process according to Claim 1, characterized in that the chromatographic separation is carried out by using a strong cation-exchanging resin as the stationary phase.

13. A process according to Claim 1, characterized in that the fermentation is carried out at a pH of about 4 - 7, preferably about 5.7, and at a temperature of about 10 - 45°C, preferably about 25 - 35°C.

14. A process according to Claim 2, characterized in that the final hydrolysis of the extracted mass is carried out enzymatically.

FIGURE: CHROMATOGRAPHIC SEPARATION



INTERNATIONAL SEARCH REPORT

International Application No PCT/FI 91/00011

I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all)[6]

According to International Patent Classification (IPC) or to both National Classification and IPC

IPC5: C 12 P 7/18, C 12 N 1/22

II. FIELDS SEARCHED

Minimum Documentation Searched[7]

Classification System	Classification Symbols
IPC5	C 12 P; C 12 N

Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in Fields Searched[8]

SE,DK,FI,NO classes as above

III. DOCUMENTS CONSIDERED TO BE RELEVANT[9]

Category *	Citation of Document,[11] with indication, where appropriate, of the relevant passages[12]	Relevant to Claim No.[13]
A	FR, A1, 2641545 (AGROCINQ) 13 July 1990, see particularly pages 3, 4, 6 --	1,7-8, 13
A	Chemical Abstracts, volume 105, no. 5, 4 August 1986, (Columbus, Ohio, US), J.C. Du Preez et al.: "Xylose fermentation by Candida shehatae and Pichia stipitis: effects of pH, temperature and substrate concentration ", see page 604, abstract 41196y, & Enzyme Microb. Technol. 1986, 8(6), 360- 364 --	1-2,7,8, 13
A	Chemical Abstracts, volume 112, no. 5, 29 January 1990, (Columbus, Ohio, US), M.T. Amaral-Collaco et al.: "Utilization of the hemicellulosic fraction of agro-industrial residues by yeasts ", see page 449, abstract 34371t, & Enzyme Syst. Lignocellul. Degrad. 1989, (), 221- 230 --	1-3,7,9

* Special categories of cited documents:[10]

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance, the claimed invention cannot be considered novel or cannot be considered to involve an inventive step

"Y" document of particular relevance, the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

IV. CERTIFICATION

Date of the Actual Completion of the International Search	Date of Mailing of this International Search Report
11th April 1991	1991 -04- 17
International Searching Authority	**Signature of Authorized Officer**
SWEDISH PATENT OFFICE	Eva Johansson

III. DOCUMENTS CONSIDERED TO BE RELEVANT (CONTINUED FROM THE SECOND SHEET)

Category *	Citation of Document, with indication, where appropriate, of the relevant passages	Relevant to Claim No
A	Chemical Abstracts, volume 114, no. 5, 4 February 1991, (Columbus, Ohio, US), K.B. Taylor et al.: "The fermentation of xylose: studies by carbon-13 nuclear magnetic resonance spectroscopy ", see page 592, abstract 41014y, & J. Ind. Microbiol. 1990, 6(1), 29- 41 --	1-2,7-8
A	Chemical Abstracts, volume 98, no. 9, 28 February 1983, (Columbus, Ohio, US), Gong, Cheng Shung et al.: "Conversion of pentoses by yeasts ", see page 484, abstract 70314c, & Biotecnol. Bioeng. 1983, 25(1), 85- 102 --	1-2,7-8

ANNEX TO THE INTERNATIONAL SEARCH REPORT ON INTERNATIONAL PATENT APPLICATION NO.PCT/FI 91/00011

This annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report.
The members are as contained in the Swedish Patent Office EDP file on 91-03-23
The Swedish Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
FR-A1- 2641545	90-07-13	NONE	

08 Rec'd PCT/FI 01 1991 AUG

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INTERNATIONAL APPLICATION UNDER THE PATENT COOPERATION TREATY

REQUEST

THE UNDERSIGNED REQUESTS THAT THE PRESENT INTERNATIONAL APPLICATION BE PROCESSED ACCORDING TO THE PATENT COOPERATION TREATY

(The following is to be filled in by the receiving Office)

INTERNATIONAL APPLICATION No: PCT/FI 9 1 / 0 0 0 1 1

INTERNATIONAL FILING DATE: 10 JAN 1991 (10 -01- 1991)

The Finnish Patent Office

(Stamp) PCT International Application

Name of receiving Office and "PCT International Application"

Applicant's or Agent's File Reference (indicated by applicant if desired) 41129/PCT/her

Box No. I TITLE OF INVENTION

A process for the simultaneous production of xylitol and ethanol

Box No. II APPLICANT (WHETHER OR NOT ALSO INVENTOR); DESIGNATED STATES FOR WHICH HE/SHE/IT IS APPLICANT. Use this box for indicating the applicant or, if there are several applicants, one of them. If more than one person (includes, where applicable, a legal entity) is involved, continue in Box No. III.

The person identified in this box is (check one only): [ ] applicant and inventor* [X] applicant only

Name *and* address:**

SUOMEN XYROFIN OY
Sokeritehtaantie
SF-48210 Kotka
Finland

Telephone number: (including area code)

Telegraphic address:

Teleprinter address:

Country of nationality: FI

Country of residence:*** FI

The person identified in this box is *applicant* for the purposes of (check one only):

[ ] all designated States [X] all designated States except the United States of America [ ] the United States of America only [ ] the States indicated in the "Supplemental Box"

Box No. III FURTHER APPLICANTS, IF ANY; (FURTHER) INVENTORS, IF ANY; DESIGNATED STATES FOR WHICH THEY ARE APPLICANTS (IF APPLICABLE). A separate sub-box has to be filled in in respect of each person (includes, where applicable, a legal entity). If the following two sub-boxes are insufficient, continue in the "Supplemental Box," (giving there for each additional person the same indications as those requested in the following two sub-boxes) or by using a "continuation sheet."

The person identified in this sub-box is (check one only): [X] applicant and inventor* [ ] applicant only [ ] inventor only*

Name *and* address:**

HEIKKILÄ Heikki
Aallonkohina 4 C 27
SF-02320 Espoo
Finland

If the person identified in this sub-box is *applicant (or applicant and inventor)*, indicate also:

Country of nationality: FI

Country of residence:*** FI

and whether that person is *applicant* for the purposes of (check one only):

[ ] all designated States [ ] all designated States except the United States of America [X] the United States of America only [ ] the States indicated in the "Supplemental Box"

The person identified in this sub-box is (check one only): [X] applicant and inventor* [ ] applicant only [ ] inventor only*

Name *and* address:**

HYÖKY Göran
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Finland

If the person identified in this sub-box is *applicant (or applicant and inventor)*, indicate also:

Country of nationality: FI

Country of residence:*** FI

and whether that person is *applicant* for the purposes of (check one only):

[ ] all designated States [ ] all designated States except the United States of America [X] the United States of America only [ ] the States indicated in the "Supplemental Box"

* If the person indicated as "applicant and inventor" or as "inventor only" is not an *inventor* for the purposes of all the designated States, give the necessary indications in the "Supplemental box."

** Indicate the name of a natural person by giving his/her family name first followed by the given name(s). Indicate the name of a legal entity by its full official designation. In the address, include both the postal code (if any) and the country (name).

*** If residence is not indicated, it will be assumed that the country of residence is the same as the country indicated in the address

Box No. III CONTINUATION (IF REQUIRED) FURTHER APPLICANTS, IF ANY; (FURTHER) INVENTORS, IF ANY; DESIGNATED STATES FOR WHICH THEY ARE APPLICANTS (IF APPLICABLE). A separate sub-box has to be filled in in respect of each person (includes, where applicable, a legal entity).

The person identified in this sub-box is (check one only): [X] applicant and inventor* [ ] applicant only [ ] inventor only*

Name *and* address:**

RAHKILA Leena
Koivuviidantie 30 A 6
SF-02130 Espoo
Finland

If the person identified in this sub-box is *applicant (or applicant and inventor)*, indicate also:

Country of nationality: FI Country of residence:***FI

and whether that person is *applicant* for the purposes of (check one only):

[ ] all designated States [ ] all designated States except the United States of America [X] the United States of America only [ ] the States indicated in the "Supplemental Box"

The person identified in this sub-box is (check one only): [X] applicant and inventor* [ ] applicant only [ ] inventor only*

Name *and* address:**

SARKKI Marja-Leena
Brissaksentie 2 D 3
SF-02460 Kantvik
Finland

If the person identified in this sub-box is *applicant (or applicant and inventor)*, indicate also:

Country of nationality: FI Country of residence:*** FI

and whether that person is *applicant* for the purposes of (check one only):

[ ] all designated States [ ] all designated States except the United States of America [X] the United States of America only [ ] the States indicated in the "Supplemental Box"

The person identified in this sub-box is (check one only): [X] applicant and inventor* [ ] applicant only [ ] inventor only*

Name *and* address:**

VILJAVA Tapio
Brontie 2, As. 12
SF-02400 Kirkkonummi
Finland

If the person identified in this sub-box is *applicant (or applicant and inventor)*, indicate also:

Country of nationality: FI Country of residence:***FI

and whether that person is *applicant* for the purposes of (check one only):

[ ] all designated States [ ] all designated States except the United States of America [X] the United States of America only [ ] the States indicated in the "Supplemental Box"

The person identified in this sub-box is (check one only): [ ] applicant and inventor* [ ] applicant only [ ] inventor only*

Name *and* address:**

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Country of nationality: Country of residence:***

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*** If residence is not indicated, it will be assumed that the country of residence is the same as the country indicated in the address.

If this continuation sheet is not used, it need not be included in the Request.

PCT/FI 91/00011

Box No. IV AGENT (IF ANY) OR COMMON REPRESENTATIVE (IF ANY); ADDRESS FOR NOTIFICATIONS (IN CERTAIN CASES). A common representative may be appointed only if there are several applicants and if no agent is or has been appointed; the common representative must be one of the applicants.
The following person (includes, where applicable, a legal entity) is hereby/has been appointed as agent or common representative to act on behalf of the applicant(s) before the competent International Authorities:

Name *and* address, including postal code and country:

OY KOLSTER AB
Stora Robertsgatan 23
P.O. Box 148
SF-00121 Helsinki
Finland

If the space below is used instead for an address for notifications, mark here ☐

Telephone number: (including area code) 358-0-618821 Telegraphic address: KOLSTERS Teleprinter address: 12-2323 KOPAT SF

Box No. V DESIGNATION OF GROUPS OF STATES OR STATES (1); CHOICE OF CERTAIN KINDS OF PROTECTION OR TREATMENT. The following designations are hereby made (please mark the applicable check-boxes):

Regional Patent

[X] EP **European Patent**(2): AT Austria, BE Belgium, CH and LI Switzerland and Liechtenstein, DE Germany (Federal Republic of), DK Denmark, ES Spain, FR France, GB United Kingdom, IT Italy, LU Luxembourg, NL Netherlands, SE Sweden, GR Greece
and any other State which is a Contracting State of the European Patent Convention and of the PCT

[ ] OA **OAPI Patent:** Benin, Burkina Faso, Cameroon, Central African Republic, Chad, Congo, Gabon, Mali, Mauritania, Senegal, Togo,
and any other State which is a Contracting State of OAPI and of the PCT; if other OAPI title desired, specify on dotted line(3):

.....

National Patent (if other kind of protection or treatment desired, specify on dotted line(3))

[ ] AT Austria(3) .....
[ ] AU Australia(3) .....
[ ] BB Barbados
[ ] BG Bulgaria(3) .....
[X] BR Brazil(3) .....
[ ] CA Canada
[ ] CH and LI Switzerland and Liechtenstein
[X] DE Germany (Federal Republic of)(3) .....
.....
[ ] DK Denmark
[ ] ES Spain(3) .....
[ ] FI Finland
[X] GB United Kingdom
[ ] HU Hungary
[X] JP Japan(3) .....
[ ] KP Democratic People's Republic of Korea(3) .....
.....
[ ] KR Republic of Korea(3) .....
[ ] LK Sri Lanka
[ ] LU Luxembourg(3) .....
[ ] MC Monaco(3) .....
[ ] MG Madagascar
[ ] MW Malawi(3) .....
[ ] NL Netherlands
[ ] NO Norway
[ ] RO Romania
[ ] SD Sudan
[ ] SE Sweden
[ ] SU Soviet Union(3) .....
.....
[X] US United States of America(3) .....
.....

Space reserved for designating States (for the purposes of a national patent) which have become party to the PCT after the issuance of this sheet:

.....

(1) The applicant's choice of the order of designations may be indicated by marking the check-boxes with sequential arabic numerals (see also the "Notes to Box No. V").
(2) The selection of particular States for a European patent can be made upon entering the national (regional) phase before the European Patent Office (see also the "Notes to Box No. V").
(3) If another kind of protection or a title of addition or, in the United States of America, treatment as a continuation or a continuation-in-part is desired, specify according to the instructions given in the "Notes to Box No. V."

PCT/FI 9 1 / 0 0 0 1 1

Box No. VI PRIORITY (IF ANY) The priority of the following earlier application is hereby claimed

Country (country in which it was filed if national application, one of the countries for which it was filed if regional or international application)	Filing Date (day, month, year)	Application No	Office of Filing (fill in only if the earlier application is an international application or a regional application)
(1) Finland	(15.1.1990) 15 January 1990	900220	
(2)			
(3)			

(Letter codes may be used to indicate country and/or Office of filing.)

When the earlier application was filed with the Office which, for the purposes of the present international application, is the receiving Office, the applicant may, *against payment of the required fee*, ask the following:

☐ the receiving Office is hereby requested to prepare and transmit to the International Bureau a certified copy of the above-mentioned earlier application/of the earlier applications identified above by the numbers (insert the applicable numbers)

Box No. VII EARLIER SEARCH (IF ANY). Fill in where a search (international, international-type or other) by the International Searching Authority has already been requested (or completed) and the said Authority is now requested to base the international search, to the extent possible, on the results of the said earlier search. Identify such search or request either by reference to the relevant application (or the translation thereof) or by reference to the search request.

International application number or number and country (or regional Office) of other application:

International/regional/national filing date

Date of request for search:

Number (if available) given to search request:

Box No. VIII SIGNATURE OF APPLICANT(S) OR AGENT

OY KOLSTER AB

by

Knut Feiring

If the present Request form is signed on behalf of any applicant by an agent, a separate power of attorney appointing the agent and signed by the applicant is required. If in such case it is desired to make use of a general power of attorney (deposited with the receiving Office), a copy thereof must be attached to this form.

Box No.IX CHECK LIST (To be filled in by the Applicant)

This international application contains the following number of sheets:

1. request	4	sheets	
2. description	22	sheets	
3. claims	2	sheets	
4. abstract	1	sheets	
5. drawings	1	sheets	
Total	30	sheets	

Figure number ..... of the drawings (if any) is suggested to accompany the abstract for publication.

This international application as filed is accompanied by the items checked below:

1. [X] separate signed power of attorney
2. [ ] copy of general power of attorney
3. [X] priority document(s) (see Box No. VI)
4. [ ] receipt of the fees paid or revenue stamps
5. [X] cheque for the payment of fees
6. [ ] request to charge deposit account
7. [X] other document (specify) Copy of Official Action Assignment Copy

(The following is to be filled in by the receiving Office)

1. Date of actual receipt of the purported international application: 10 -01- 1991
2. Corrected date of actual receipt due to later but timely received papers or drawings completing the purported international application:
3. Date of timely receipt of the required corrections under Article 11 of the PCT:
4. Drawings [ ] Received [ ] No Drawings

(The following is to be filled in by the International Bureau)

Date of receipt of the record copy: 28 JANUARY 1991 (28. 01. 91)

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INTERNATIONAL APPLICATION UNDER THE PATENT COOPERATION TREATY

REQUEST

THE UNDERSIGNED REQUESTS THAT THE PRESENT INTERNATIONAL APPLICATION BE PROCESSED ACCORDING TO THE PATENT COOPERATION TREATY

(The following is to be filled in by the receiving Office)

INTERNATIONAL APPLICATION No: PCT/FI 91/00011

INTERNATIONAL FILING DATE: 10 JAN 1991 (10-01-1991)

The Finnish Patent Office

(Stamp) PCT International Application

Name of receiving Office and "PCT International Application"

Applicant's or Agent's File Reference (indicated by applicant if desired) 41129/PCT/her

Box No. I TITLE OF INVENTION

A process for the simultaneous production of xylitol and ethanol

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The person identified in this box is (check one only): [ ] applicant and inventor* [X] applicant only

Name *and* address:**

SUOMEN XYROFIN OY
Sokeritehtaantie
SF-48210 Kotka
Finland

Telephone number: (including area code)

Telegraphic address:

Teleprinter address:

Country of nationality: FI

Country of residence:*** FI

The person identified in this box is *applicant* for the purposes of (check one only):

[ ] all designated States [X] all designated States except the United States of America [ ] the United States of America only [ ] the States indicated in the "Supplemental Box"

Box No. III FURTHER APPLICANTS, IF ANY; (FURTHER) INVENTORS, IF ANY; DESIGNATED STATES FOR WHICH THEY ARE APPLICANTS (IF APPLICABLE). A separate sub-box has to be filled in in respect of each person (includes, where applicable, a legal entity). If the following two sub-boxes are insufficient, continue in the "Supplemental Box," (giving there for each additional person the same indications as those requested in the following two sub-boxes) or by using a "continuation sheet."

The person identified in this sub-box is (check one only): [X] applicant and inventor* [ ] applicant only [ ] inventor only*

Name *and* address:**

HEIKKILÄ Heikki
Aallonkohina 4 C 27
SF-02320 Espoo
Finland

If the person identified in this sub-box is *applicant (or applicant and inventor)*, indicate also:

Country of nationality: FI

Country of residence:*** FI

and whether that person is *applicant* for the purposes of (check one only):

[ ] all designated States [ ] all designated States except the United States of America [X] the United States of America only [ ] the States indicated in the "Supplemental Box"

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Name *and* address:**

HYÖKY Göran
Linkoojanrinne 7
SF-02460 Kantvik
Finland

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Country of nationality: FI

Country of residence:*** FI

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** Indicate the name of a natural person by giving his/her family name first followed by the given name(s). Indicate the name of a legal entity by its full official designation. In the address, include both the postal code (if any) and the country (name).

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PCT/FI 9 1 / 0 0 0 11

Box No. III CONTINUATION (IF REQUIRED) FURTHER APPLICANTS, IF ANY; (FURTHER) INVENTORS, IF ANY; DESIGNATED STATES FOR WHICH THEY ARE APPLICANTS (IF APPLICABLE). A separate sub-box has to be filled in in respect of each person (includes, where applicable, a legal entity).

The person identified in this sub-box is (check one only): [X] applicant and inventor* [ ] applicant only [ ] inventor only*

Name *and* address:**

RAHKILA Leena
Koivuviidantie 30 A 6
SF-02130 Espoo
Finland

If the person identified in this sub-box is *applicant (or applicant and inventor)*, indicate also:

Country of nationality: FI Country of residence:***FI

and whether that person is *applicant* for the purposes of (check one only):

[ ] all designated States [ ] all designated States except the United States of America [X] the United States of America only [ ] the States indicated in the "Supplemental Box"

The person identified in this sub-box is (check one only): [X] applicant and inventor* [ ] applicant only [ ] inventor only*

Name *and* address:**

SARKKI Marja-Leena
Brissaksentie 2 D 3
SF-02460 Kantvik
Finland

If the person identified in this sub-box is *applicant (or applicant and inventor)*, indicate also:

Country of nationality: FI Country of residence:*** FI

and whether that person is *applicant* for the purposes of (check one only):

[ ] all designated States [ ] all designated States except the United States of America [X] the United States of America only [ ] the States indicated in the "Supplemental Box"

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Name *and* address:**

VILJAVA Tapio
Brontie 2, As. 12
SF-02400 Kirkkonummi
Finland

If the person identified in this sub-box is *applicant (or applicant and inventor)*, indicate also:

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The person identified in this sub-box is (check one only): [ ] applicant and inventor* [ ] applicant only [ ] inventor only*

Name *and* address:**

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and whether that person is *applicant* for the purposes of (check one only):

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If this continuation sheet is not used, it need not be included in the Request.

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The following person (includes, where applicable, a legal entity) is hereby/has been appointed as agent or common representative to act on behalf of the applicant(s) before the competent International Authorities

Name *and* address, including postal code and country

OY KOLSTER AB
Stora Robertsgatan 23
P.O. Box 148
SF-00121 Helsinki
Finland

If the space below is used instead for an address for notifications, mark here ☐

Telephone number: (including area code) 358-0-618821 Telegraphic address: KOLSTERS Teleprinter address: 12-2323 KOPAT SF

Box No. V DESIGNATION OF GROUPS OF STATES OR STATES (1); CHOICE OF CERTAIN KINDS OF PROTECTION OR TREATMENT. The following designations are hereby made (please mark the applicable check-boxes):

Regional Patent

[X] EP European Patent(2): AT Austria, BE Belgium, CH and LI Switzerland and Liechtenstein, DE Germany (Federal Republic of), DK Denmark, ES Spain, FR France, GB United Kingdom, IT Italy, LU Luxembourg, NL Netherlands, SE Sweden, GR Greece
and any other State which is a Contracting State of the European Patent Convention and of the PCT

[ ] OA OAPI Patent: Benin, Burkina Faso, Cameroon, Central African Republic, Chad, Congo, Gabon, Mali, Mauritania, Senegal, Togo,
and any other State which is a Contracting State of OAPI and of the PCT; if other OAPI title desired, specify on dotted line(3): .....

National Patent (if other kind of protection or treatment desired, specify on dotted line(3))

[ ] AT Austria(3) .....
[ ] AU Australia(3) .....
[ ] BB Barbados
[ ] BG Bulgaria(3) .....
[X] BR Brazil(3) .....
[ ] CA Canada
[ ] CH and LI Switzerland and Liechtenstein
[X] DE Germany (Federal Republic of)(3) .....
.....
[ ] DK Denmark
[ ] ES Spain(3) .....
[ ] FI Finland
[X] GB United Kingdom
[ ] HU Hungary
[X] JP Japan(3) .....
[ ] KP Democratic People's Republic of Korea(3) .....
.....
[ ] KR Republic of Korea(3) .....
[ ] LK Sri Lanka
[ ] LU Luxembourg(3) .....
[ ] MC Monaco(3) .....
[ ] MG Madagascar
[ ] MW Malawi(3) .....
[ ] NL Netherlands
[ ] NO Norway
[ ] RO Romania
[ ] SD Sudan
[ ] SE Sweden
[ ] SU Soviet Union(3) .....
.....
[X] US United States of America(3) .....
.....

Space reserved for designating States (for the purposes of a national patent) which have become party to the PCT after the issuance of this sheet:

.....

(1) The applicant's choice of the order of designations may be indicated by marking the check-boxes with sequential arabic numerals (see also the "Notes to Box No. V").
(2) The selection of particular States for a European patent can be made upon entering the national (regional) phase before the European Patent Office (see also the "Notes to Box No. V").
(3) If another kind of protection or a title of addition or, in the United States of America, treatment as a continuation or a continuation-in-part is desired, specify according to the instructions given in the "Notes to Box No. V"

Box No. VI PRIORITY [illegible]M (IF ANY) The priority of the following earlier [illegible]ation(s) is hereby claimed

Country (country in which it was filed if national application, one of the countries for which it was filed if regional or international application)	Filing Date (day, month, year)	Application No	Office of Filing (fill in only if the earlier application is an international application or a regional application)
(1) Finland	(15.1.1990) 15 January 1990	900220	
(2)			
(3)			

(Letter codes may be used to indicate country and/or Office of filing.)

When the earlier application was filed with the Office which, for the purposes of the present international application, is the receiving Office, the applicant may, *against payment of the required fee*, ask the following.

☐ the receiving Office is hereby requested to prepare and transmit to the International Bureau a certified copy of the above-mentioned earlier application/of the earlier applications identified above by the numbers (insert the applicable numbers)

Box No. VII EARLIER SEARCH (IF ANY) Fill in where a search (international, international-type or other) by the International Searching Authority has already been requested (or completed) and the said Authority is now requested to base the international search, to the extent possible, on the results of the said earlier search. Identify such search or request either by reference to the relevant application (or the translation thereof) or by reference to the search request.

International application number or number and country (or regional Office) of other application:

International/regional/national filing date

Date of request for search:

Number (if available) given to search request:

Box No. VIII SIGNATURE OF APPLICANT(S) OR AGENT

OY KOLSTER AB

by

Knut Felring

If the present Request form is signed on behalf of any applicant by an agent, a separate power of attorney appointing the agent and signed by the applicant is required. If in such case it is desired to make use of a general power of attorney (deposited with the receiving Office), a copy thereof must be attached to this form.

Box No.IX CHECK LIST (To be filled in by the Applicant)

This international application contains the following number of sheets:

1. request	4	sheets	
2. description	22	sheets	
3. claims	2	sheets	
4. abstract	1	sheets	
5. drawings	1	sheets	
Total	30	sheets	

Figure number ... of the drawings (if any) is suggested to accompany the abstract for publication.

This international application as filed is accompanied by the items checked below:

1.	[X]	separate signed power of attorney
2.	[ ]	copy of general power of attorney
3.	[X]	priority document(s) (see Box No. VI)
4.	[ ]	receipt of the fees paid or revenue stamps
5.	[X]	cheque for the payment of fees
6.	[ ]	request to charge deposit account
-	[X]	other document (specify) Copy of Official Action Assignment Copy

(The following is to be filled in by the receiving Office)

1. Date of actual receipt of the purported international application: 10 -01- 1991
2. Corrected date of actual receipt due to later but timely received papers or drawings completing the purported international application:
3. Date of timely receipt of the required corrections under Article 11 of the PCT:
4. Drawings [ ] Received [ ] No Drawings

(The following is to be filled in by the International Bureau)

Date of receipt of the record copy:

6

PATENT COOPERATION TREATY
INTERNATIONAL PRELIMINARY EXAMINATION REPORT

IDENTIFICATION OF THE INTERNATIONAL APPLICATION	Applicant's or Agent's File Reference
	41129/PCT/her
International Application No. PCT/FI91/00011	International Filing Date 1991-01-10
Receiving Office RO/FI	Priority Date Claimed 1990-01-15
Applicant (Name) SUOMEN XYROFIN OY et al	

BASIS OF REPORT

1. AMENDMENTS AND/OR RECTIFICATIONS[1]* — The amendments and/or rectifications made before this International Preliminary Examining Authority in respect of the claims, the description, and/or drawings in the above-identified international application are annexed to this report.

a. [X] This report has been established on the basis of the following application documents:

[X] the application documents as filed		1991-01-10
[ ] description, pages	as originally filed	.....
description, pages	filed with your letter of	.....
description, pages	filed with your letter of	.....
description, pages	filed with your letter of	.....
[ ] claim(s)	as originally filed	.....
claim(s)	filed with your letter of	.....
claim(s)	filed with your letter of	.....
claim(s)	filed with your letter of	.....
[ ] drawings, sheet fig.	as originally filed	.....
drawings, sheet fig.	filed with your letter of	.....

b. [ ] The amendments resulted in the cancellation of the following sheets: .....

c. [ ] This report has been established as if the amendments indicated on the extra sheet have not been made, since, for the reasons indicated, they have been considered to go beyond the disclosure as filed.

2. PRIORITY[2]

a. This report has been established as if no priority has been claimed due to the failure to furnish within the prescribed time limit the requested:

[ ] copy of the earlier application whose priority has been claimed.

[ ] translation of the earlier application whose priority has been claimed.

b. [ ] This report has been established as if no priority has been claimed due to the fact that the priority claim has been found invalid.

Thus, for the purposes of this report, the international filing date indicated above is considered to be the relevant date.

* Where replacement sheets are annexed to this report, a translation of these replacement sheets must be furnished to the elected Offices within the time limit applicable under PCT Article 39(1).

See notes on accompanying sheet

CLASSIFICATION OF SUBJECT MATTER (If several classification symbols apply, indicate all.) [6]

According to International Patent Classification (IPC) or to both National Classification and IPC 5

C 12 P 7/18, C 12 N 1/22

REASONED STATEMENT AS TO CLAIMS MEETING CRITERIA OF NOVELTY (N), INVENTIVE STEP (IS) AND INDUSTRIAL APPLICABILITY (IA) [6] **AND CITATIONS** [7] **AND EXPLANATIONS** [8] **SUPPORTING SUCH STATEMENT**

CLAIM NUMBER	STATEMENT (CRITERIA)	CITATIONS AND EXPLANATIONS
1-14	Yes (N, IS, IA)	The present invention relates to a process for the simultaneous production of xylitol and ethanol. A hydrolyzed lignocellulose-containing material is fermented with a yeast strain. The ethanol is recovered and then pure xylitol is obtained after a chromatographic separation. The cited documents only provide prior art which is not relevant to the subject matter of the present claims. Therefore, the claimed invention is considered to fulfil the requirements of novelty, inventive step and industrial applicability.

NON-WRITTEN DISCLOSURES [13]

Kind of Non-Written Disclosure	Date of Written Disclosure referring to the Non-Written Disclosure	Date of Non-Written Disclosure

CERTAIN PUBLISHED DOCUMENTS [14]

Application/Patent	Date of Publication	Filing Date	Priority Date (Valid Claim)

CERTAIN DEFECTS IN THE INTERNATIONAL APPLICATION [15]

The following defects in the form or contents of the international application have been noted.

CERTAIN OBSERVATIONS ON THE INTERNATIONAL APPLICATION [16]

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description have been noted.

CERTIFICATION

Date Demand Submitted [17]	Date of Completion of the International Preliminary Examination Report [18]
1991-06-05	1992-04-10
International Preliminary Examining Authority [1]	**Signature of Authorized Officer** [19]
Swedish Patent Office	Anna Sjölund

See notes on accompanying sheet

PATENT COOPERATION TREATY

INTERNATIONAL APPLICATION No. PCT/FI91/00011

INFORMATION CONCERNING ELECTED OFFICES NOTIFIED OF THEIR ELECTION
issued pursuant to PCT Rule 61.3

To:

OY KOLSTER AB
Stora Robertsgatan 23
P.O. Box 148
SF-00121 Helsinki
FINLANDE

DATE OF MAILING OF THIS NOTIFICATION:
31 July 1991 (31.07.91)

APPLICANT'S OR AGENT'S FILE REFERENCE:
41129/PCT/her

From:
The International Bureau of WIPO
1211 Geneva 20
Switzerland

APPLICANT (NAME):
SUOMEN XYROFIN OY

INTERNATIONAL FILING DATE:
10 January 1991 (10.01.91)

The International Bureau has notified, as provided in PCT Article 31(7), each of the following Offices of its election:
NATIONAL OFFICES OF: BR,DE,GB,JP,US
EP (for AT,BE,DE,DK,FR,GB,IT,LU,NL,SE)

The applicant is reminded that he must enter the "national phase" before the expiration of 30 months from the priority date before each of the Offices listed above, provided that the relevant election took place prior to the expiration of the 19th month from the priority date. This must be done by performing the acts referred to in PCT Article 39(1)(a) (i.e., paying the national fee(s) and furnishing, if prescribed, a translation of the international application), as well as, where applicable, under PCT Article 36(3)(b) and PCT Rule 74.1, by furnishing a translation of any annex of the international preliminary examination report.

The following Offices have fixed time limits which expire later:
- AU, BG and EP: 31 months from the priority date;
- RO: 35 months from the priority date;
- PL: a translation into Polish must always be furnished within 20 months from the priority date, even where Poland was elected for international preliminary examination before the expiry of 19 months from the priority date.

For detailed information about the acts to be performed and the applicable time limits, see Volume II of the PCT Applicant's Guide.

Form PCT/IB/332 (June 1991)

M.C. Taylor
(Authorized Officer)

PATENT COOPERATION TREATY

25-07-1991

TO

OY KOLSTER AB
Stora Robertsgatan 23
P.O. Box 148
SF-00121 HELSINKI
FINLAND

FROM the INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY identified at the bottom of this page

NOTIFICATION OF RECEIPT OF DEMAND
issued pursuant to PCT Rule 61.1(b), first sentence

Inscribe NAME and ADDRESS of the AGENT and if there is no agent, of the APPLICANT

DATE OF MAILING by the International Preliminary Examining Authority: 1991 -07- 23

APPLICANT'S OR AGENT'S FILE REFERENCE: 41129/PCT/her

IDENTIFICATION OF THE INTERNATIONAL APPLICATION

International Application No.	International Filing Date
PCT/FI91/00011	1991-01-10

Applicant (Name)

SUOMEN XYROFIN OY et al

NOTIFICATION

The applicant is hereby notified that this International Preliminary Examining Authority considers the following date as the date of receipt of the demand for international preliminary examination of the above-identified international application:

1991-06-05
(date of receipt)

This date of receipt corresponds with the following indicated date:

1. [X] the actual date of receipt of the demand.

2. [ ] the date on which the proper corrections to the demand were timely received.

THE INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

Name and Mailing Address	Telephone	Authorized Officer
Patent- och registreringsverket Box 5055 S-102 42 STOCKHOLM	08-782 25 00 Telex 17978 PATOREG-S	Lotta Hjelmersson

Form PCT/IPEA/402 (January 1985)

PATENT COOPERATION TREATY

DEMAND

UNDER ARTICLE 31 OF THE PATENT COOPERATION TREATY:

THE UNDERSIGNED REQUESTS THAT THE INTERNATIONAL APPLICATION SPECIFIED BELOW BE THE SUBJECT OF INTERNATIONAL PRELIMINARY EXAMINATION ACCORDING TO THE PATENT COOPERATION TREATY

CHAPTER II

Box No. I IDENTIFICATION OF THE INTERNATIONAL APPLICATION		Applicant's or Agent's File Reference (indicated by applicant if desired):
International Application No. PCT/FI91/00011	International Filing Date 10 January 1991	(Earliest) Priority Date 15 January 1990

Title of Invention

A process for the simultaneous production of xylitol and ethanol

Box No. II APPLICANT(S). Further applicants are indicated on a continuation sheet [X]

Name *and* address, including postal code and country:

SUOMEN XYROFIN OY
Sokeritehtaantie
SF-48210 Kotka
Finland

State of nationality: FI

State of residence:* FI

Telephone number (including area code):

Telegraphic address:

Teleprinter address:

Name *and* address, including postal code and country:

HEIKKILÄ Heikki
Aallonkohina 4 C 27
SF-02320 Espoo
Finland

State of nationality: FI

State of residence:* FI

Box No. III AGENT OR COMMON REPRESENTATIVE (IF ANY); ADDRESS FOR NOTIFICATIONS (IN CERTAIN CASES)

The following named agent or common representative

1. [x] has been appointed earlier and represents the applicant also for international preliminary examination
2. [ ] is hereby appointed and any earlier appointment of an agent is hereby revoked
3. [ ] is hereby appointed, in addition to the agent(s) appointed earlier, for the procedure before the International Preliminary Examining Authority

Name *and* address, including postal code and country:

If the space below is used instead for an address for notifications, mark here: [ ]

Telephone number (including area code):

Telegraphic address:

Teleprinter address:

* If residence is not indicated, it will be assumed that the State of residence is the same as the State indicated in the address.

Box No. IV DECLARATION CONCERNING AMENDMENTS OF THE CLAIMS

Applicant wishes international preliminary examination to start promptly on the basis of the claims

[x] as filed (amendments under Article 19 have not been made and will not be made)

[ ] as amended under Article 19

[ ] as specified on the attached sheet

Box No. V ELECTION OF STATES

The following designated States are hereby elected (please mark the applicable check-boxes):

Regional Patent

[x] EP European Patent: AT Austria, BE Belgium, DE Germany, DK Denmark, FR France, GB United Kingdom, IT Italy, LU Luxembourg, NL Netherlands, SE Sweden, and any other State which is a Contracting State of the European Patent Convention and of the PCT (including Chapter II thereof).

[ ] OA OAPI Patent: Benin, Burkina Faso, Cameroon, Central African Republic, Chad, Congo, Gabon, Mali, Mauritania, Senegal, Togo, and any other State which is a Contracting State of the OAPI and of the PCT (including Chapter II thereof).

National Patent

[ ] AT Austria
[ ] AU Australia
[ ] BB Barbados
[ ] BG Bulgaria
[x] BR Brazil
[ ] CA Canada
[x] DE Germany
[ ] DK Denmark
[ ] FI Finland
[x] GB United Kingdom
[ ] HU Hungary
[x] JP Japan
[ ] KP Democratic People's Republic of Korea
[ ] KR Republic of Korea
[ ] LK Sri Lanka
[ ] LU Luxembourg
[ ] MC Monaco
[ ] MG Madagascar
[ ] MW Malawi
[ ] NL Netherlands
[ ] NO Norway
[ ] PL Poland
[ ] RO Romania
[ ] SD Sudan
[ ] SE Sweden
[ ] SU Soviet Union
[x] US United States of America

Space reserved for electing States which have become party to the PCT (including Chapter II thereof) or bound by Chapter II of the PCT after the issuance of this sheet:

.....

Box No. VI SIGNATURE

Oy KOLSTER Ab

by

Knut Feiring

(The following is to be filled in by the International Preliminary Examining Authority)

1. Date of actual receipt of DEMAND:

2. Adjusted date of receipt of DEMAND due to CORRECTIONS under Rule 60.1(b):

Box No. II APPLICANT(S) (CON[redacted]CATION)

Name *and* address, including postal code and country:

HYÖKY Göran
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State of nationality: FI

State of residence:* FI

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State of residence:* FI

Name *and* address, including postal code and country:

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State of nationality: FI

State of residence:* FI

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Finland

State of nationality: FI

State of residence:* FI

Name *and* address, including postal code and country:

State of nationality:

State of residence:*

Name *and* address, including postal code and country:

State of nationality:

State of residence:*

* If residence is not indicated, it will be assumed that the State of residence is the same as the State indicated in the address.

International Application No: F191-11 PCT/910133

FEES SUBMITTED OR AUTHORIZED:

NOTE: Please stamp file "FILED UNDER 35 USC 371"

Non-Small Entity Patent Processing Fees

FEE	CODE	CASH	CHARGE
Basic		$ .00	$ 00
		$ 00	$ 00
$920	960	$ .00	$ 920 00 1140
		$ .00	$ .00
		$ 00	$ 00
Each Independent Claim in EXCESS OF 3		$ .00	$ .00
Claim(s) in EXCESS OF 20		$ .00	$ .00
Multiple Dependent Claims		$ .00	$ .00
Surcharge for Late Fees/Oath or Declaration	154	$ .00	$ .00

Small Entity Patent Processing Fees

FEE	CODE	CASH	CHARGE
Basic		$ 00	$ .00
		$ 00	$ .00
$460	961	$ .00	$ .00
		$ 00	$ .00
		$ 00	$ 00
Each Independent Claim in EXCESS OF 3		$ 00	$ .00
Claim(s) in EXCESS OF 20		$ 00	$ .00
Multiple Dependent Claim(s)		$ 00	$ .00
Surcharge for Late Fee/Oath or Declaration		$ 00	$ .00

Other Fees - NO SMALL ENTITY REDUCTION

	CODE	CASH	CHARGE
Surcharge for Late Trans.	156	$ 00	$ .00
Assignment	581	$ 00	$ .00

Total Cash - $ 00

Total Charge - $ 1140 00

Marja-Leena Sarkki

06.07.99

Translation from the Finnish original text:

HYDROLYSIS OF LACTOSE (WHEY) WITH IMMOBILIZED BETA – GALACTOSIDASE

THESIS OF MR. KARI HYRKÄS IN HELSINKI TECHNICAL UNIVERSITY

PUBLISHED 12 DECEMBER 1974

The activity of immobilized enzyme increases until the saturation value which is depending on the particle size of resin when the amount of enzyme used for immobilization is increased.

However the higher the enzyme concentration of immobilization liquid is the smaller the activity yield is. Thus optimization of immobilization conditions requires information on equipment and operating costs.

By utilizing the method used in the study it is easy to produce immobilized enzyme which is considerably more active than commercial Duolase L-10.

It is recommendable to immobilize enzyme with a resin as fine as possible suitable for desired use because the activity of immobilized enzyme increases while the particle size of resin decreases in the same immobilization conditions. It was observed that resin of particle size 0,25...0,35 mm suits well for both column and batch processes.

The same resin can be used at least twice for the immobilization of an enzyme.

The immobilization liquid can be reused for production of immobilized enzyme, though the yield is only 44 % of the yield of new enzyme.

According to the activity measurements the maximum activity of immobilized enzyme was at 60 °C while the reaction time of soluble enzyme was 10 minutes. The activity of enzyme in the column increased when temperature was increased, at least until 65 °C. Optimal pH –value of immobilized enzyme is narrower than the one of soluble enzyme, but the stability is better when pH –value is over 5. Eventhough optimum pH –value of immobilized enzyme is 4,0, the activity is 90 % of the maximum at pH 4,5.

Other substrate compounds than lactose cause inhibition of enzyme, which is higher with whey than with whey filtrate.

The stability of immobilized enzyme is considerably weaker at 45 °C and at 60 °C than the stability of the soluble enzyme.

The enzyme inactivates while drying almost completely. The leaking of column totally did not however inactivate the enzyme.

Marja-Leena Sarkki 06.07.99

The stability of immobilized enzyme was good during the process use. The enzyme leaked slightly in both batch and column use in the beginning, but thereafter decreasing of activity was slow. Half life of the enzyme activity was measured to be 20 .. 28 days in a continuous hydrolysis of whey at 45 °C. The results are however based only on the stability experiments of 8 days.

Insoluble protein, so called "dust" , gathered in column in continuous hydrolysis of whey, which caused weakening of function of enzyme unless "dust" was removed.

In batch hydrolysis of ultrafiltrate at 45 °C 80 % hydrolysis of lactose was reached in 4.5 hours when the proportion of immobilized enzyme/ lactose of substrate was 0,78. The corresponding hydrolysis degree was reached at 50 °C in about 3,7 hours. 100 % hydrolysis of whey's lactose at 45 °C in nine hours was reached when the proportion was 1,28.

In continuous hydrolysis of whey quite high hydrolysis degree were reached with the enzyme column by using rather high feed rates. At the feed rate, four column bed volume per hour, at 45 °C the hydrolysis degree was 80 % and 60 % when the feed rate was about eleven column bed volumes per hour.

On the basis of price estimations of enzyme and resin found in literature and on the basis of the stability tests performed in this study the use of the immobilized enzyme for hydrolysis of 80 % of whey lactose seem to be more economical than the one of soluble enzyme.

Translation July 6 th, 1999 in Kantvik , Finland

Leena Ristakoski

Marja-Leena Sarkki

CULTOR
Food Science

Marja-Leena Sarkki

03.06.99

HYDROLYSIS OF LACTOSE (WHEY) WITH IMMOBILIZED BETA-GALACTOSIDASE

THESIS OF MR. KARI HYRKÄS IN HELSINKI TECHNICAL UNIVERSITY

PUBLISHED DECEMBER 12.1974

Summary of the pages 39-47 prepared by Marja-Leena Sarkki , Cultor Oyj from the finnish text, referred in the prosecution in Finland

The referred part of the study clarifies, how much in the immobilization process had to be used beta-glucosidase enzyme in the solution to obtain a certain bounded enzyme activity (U/g resin) on a carrier. The bounded enzyme was measured as protein (Lowry- method) from the filtrate solution after immobilization of enzyme on the resin. Glutaraldehyde was used in the immobilization and therefore the filtrate solution contained glutaraldehyde, which was disturbing the Lowry method. The other method used for the estimation of the quantity of the bounded enzyme was a measurement as enzyme activity from the filtrate and from the immobilized resin. The losses of enzyme activity during immobilization were 30-50 % due to reactions with glutaraldehyde and due to the denaturation caused by glutaraldehyde. The saturation level of resin was 240 units (beta-galactosidase)/ g resin , when the amount of protein used in immobilization was 25 mg/g resin. The immobilization method used in the study produced improved carrier material compared to a commercial beta-galactosidase Duolase L-10 that contained 103 U/ g resin.
The experimental study also deals with the effect of the particle size of resin on the immobilization and reuse of carrier material and reuse of the eluate from the immobilization process.
Using glutaraldehyde in the immobilization process has carried out in all the experiments. Experiments without glutaraldehyde were speculated only for testing a theoretical adsorption of enzyme on the used resin as an analysis method not to be used for the practical process.

PATENTTI- JA REKISTERIHALLITUS

VÄLIPÄÄTÖS

30.10.90

Oy Kolster Ab

Iso Roobertinkatu 23

00120 Helsinki

Patenttihakemus nro:	900220
Luokka:	C 12P 007/02 / NL
Hakija:	Cultor Oy
Asiamies:	Oy Kolster Ab
Asiamiehen viite:	2891540FI
Määräpäivä	30.04.91

Patenttihakemuksen numero ja luokka on mainittava kirjelmässänne PRH:lle

---

Vaatimuksen 1 mukaisen menetelmän tekniikan tasona mainitaan US-patenttijulkaisu 4 368 268 (erit. palsta 1 rivit 17-23) sekä julkaisut Process Biochemistry 24(1) (1989) 21-32 (erit. s. 22 rivit 1-6 ja s. 23 rivit 8-16) ja Enzyme and Microbial Technology 9(1) (1987) 5-15 (erit. s. 8 toinen palsta rivit 13-17). Nämä muodostavat esteen nykyisen epätäsmällisesti määritellyn vaatimuksen 1 mukaisen menetelmän patentoimiselle (PL 2 § 1 mom.). Hakijan tiedoksi lähetetään myös kopio julkaisusta Biotechnology Letters 12(1) (1990) 57-60, joka ei kuitenkaan ole este hakemukselle.

Patenttivaatimus 1 on epätäsmällinen ja liian laaja siihen nähden, mitä esimerkkien nojalla on perusteltavissa (PL 8 § 2 mom. ja PM 5 § 2). Käytetty hiivakanta tulee täsmentää. Vaatimuksessa 5 esitetyt käytetyn hiivakannan ominaisuudet ovat välttämättömät vaatimuksen 1 mukaisen menetelmän suorittamiseksi, joten vaatimus 5 tulee sisällyttää vaatimukseen 1.

Patenttivaatimukset 2 ja 13 eivät kuvaa vaatimuksen 1 mukaista menetelmää, koska fermentaatio suoritetaan kahdessa eri vaiheessa eikä samanaikaisesti. Myöskään esimerkki 8 ei kuvaa vaatimusten mukaista menetelmää, koska siinä tuotetaan ainoastaan etanolia.

Nina Lindholm

Tutkijainsinööri
Puhelin: (90) 6953347
1537e/ESO

Nina Lindholm

Lausumanne huomautusten johdosta on annettava viimeistään yllämainittuna määräpäivänä. Jollette ole antanut lausumaanne virastoon viimeistään mainittuna määräpäivänä tai ryhtynyt toimenpiteisiin tässä välipäätöksessä esitettyjen puutteellisuuksien korjaamiseksi, jätetään hakemus sillensä (patenttilain 15 §). Sillensä jätetty hakemus otetaan uudelleen käsiteltäväksi, jos Te neljän kuukauden kuluessa määräpäivästä annatte lausumanne tai ryhdytte toimenpiteisiin esitettyjen puutteellisuuksien korjaamiseksi ja samassa ajassa suoritatte vahvistetun maksun, 280 mk hakemuksen ottamisesta uudelleen käsiteltäväksi. Jos lausumanne on annettu virastoon oikeassa ajassa, mutta esitettyjä puutteellisuuksia ei ole siten korjattu, että hakemus voitaisiin hyväksyä, se hylätään, mikäli virastolla ei ole aihetta antaa Teille uutta välipäätöstä (patenttilain 16 §). Uusi keksinnön selitys, siihen tehdyt lisäykset ja uudet patenttivaatimukset on aina jätettävä kahtena kappaleena.

Postiosoite: Pl 154, 00181 Helsinki
Katuosoite: Albertinkatu 25, 00180 Helsinki
Puhelin: (90) 69531
Telefax: (90) 6953204
Pankki: ps 4790-8

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